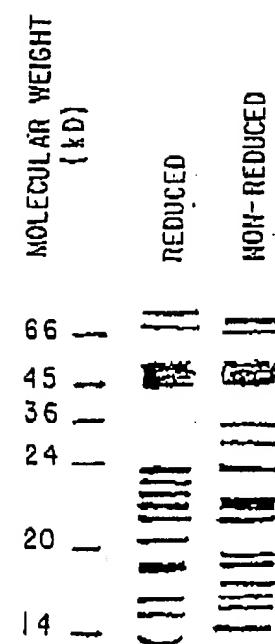


G022077 8200000000

FIGURE 1



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FIGURE ■ 2

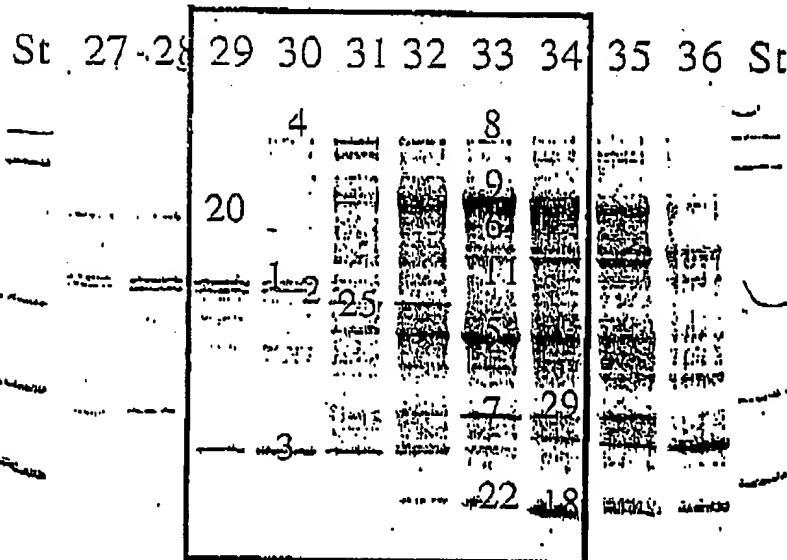
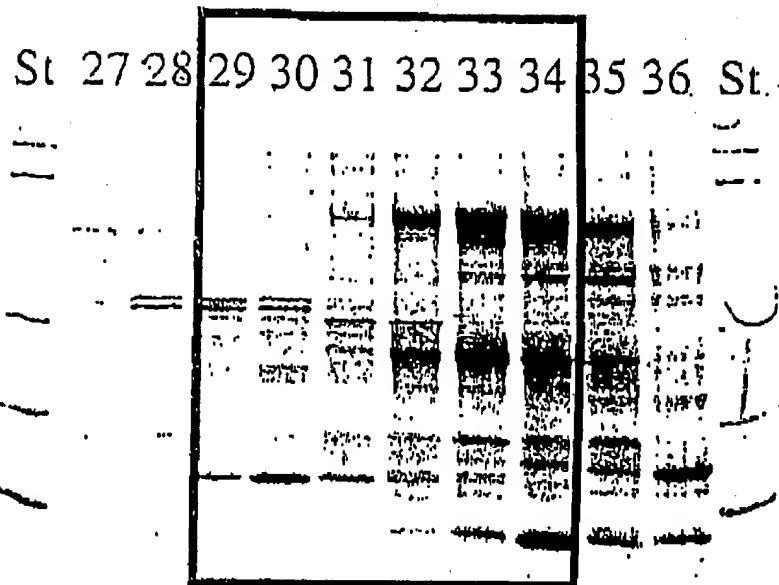


FIGURE ■ 3

00222247-2202-4745-9

00000000000000000000000000000000



Band No.	Identity
1	histone H1.c
2	histone H1.c
3	ribosomal protein RS20
4	similar to ribosomal protein LORP
5	BMP-3
6	α 2 macroglobulin RAP & BMP-3
7	similar to ribosomal protein LORP
8	BMP-3
9	BMP-3
11	ribosomal protein RL6 & BMP-3
18	TGF- β 2/SPP24
20	Factor H
22	TGF- β 2
25	BMP-3 & H1.x
29	BMP-3 & ribosomal protein RL32

FIGURE 4

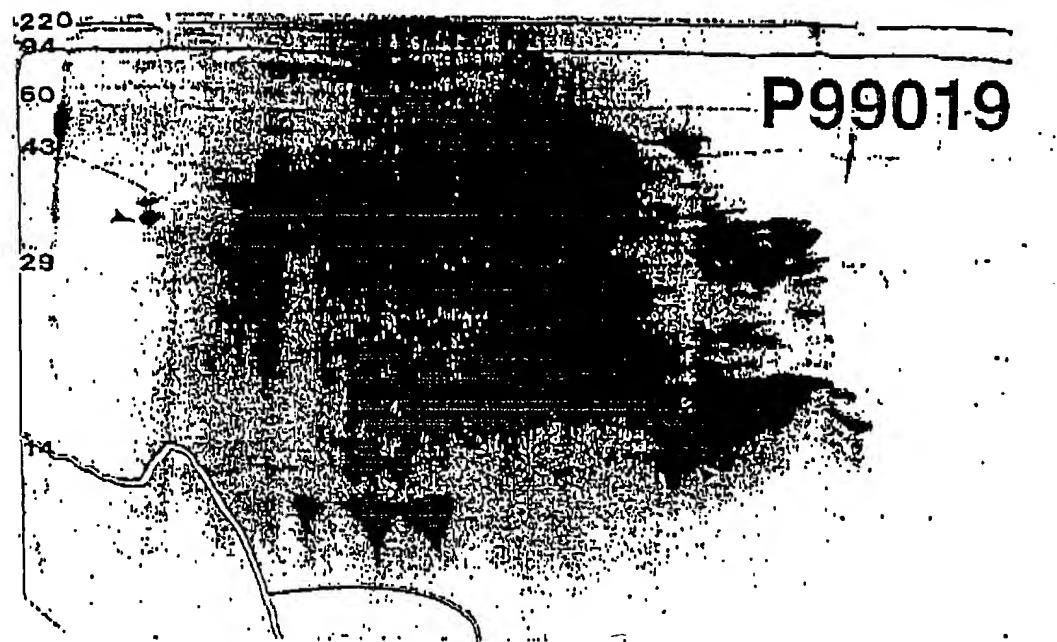


FIGURE 5

Proteinase XIIIb
Proteinase XIIIa
Cathepsin L pre.
Cathepsin L Oxidase
SPP-4/TGF- β 2
SPP-4
SPP-4/TGF β 2
Chromosome L3
Chromosome S3a
Chromosome H1.c
Chromosome S4

Figure 6

Figure 3A. (Band 1)

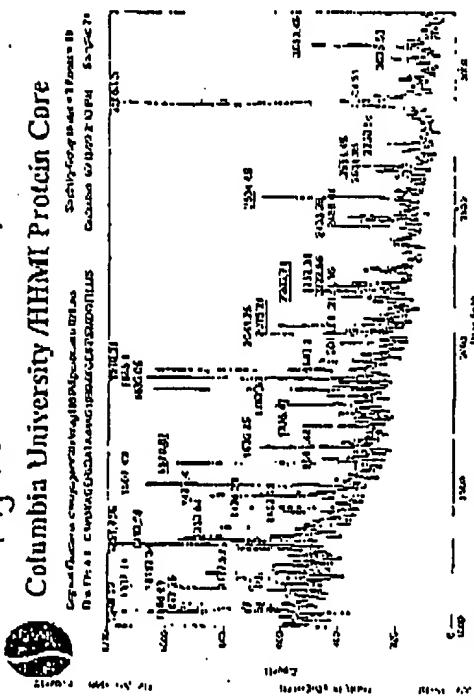
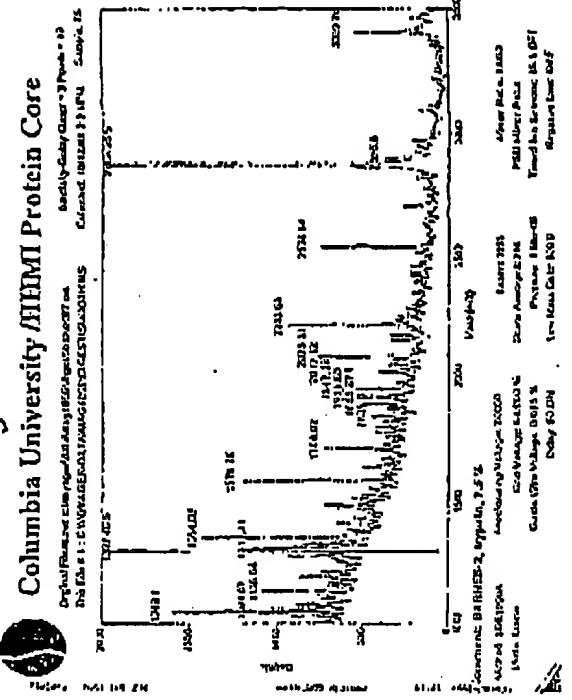
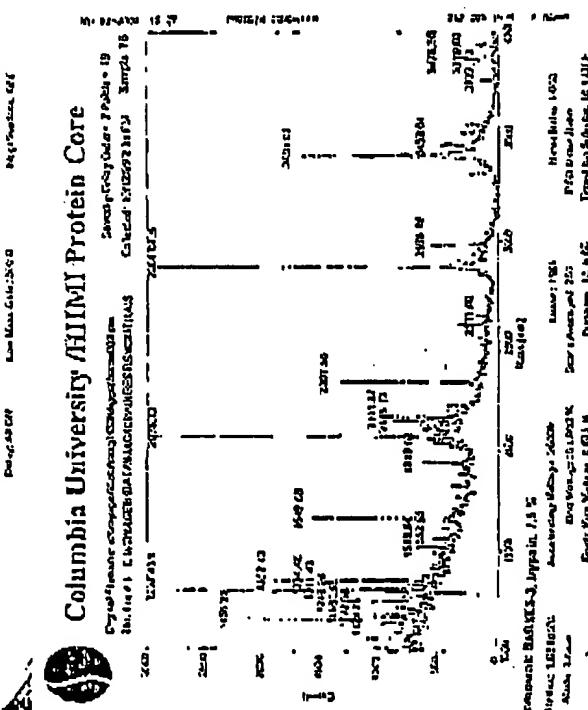


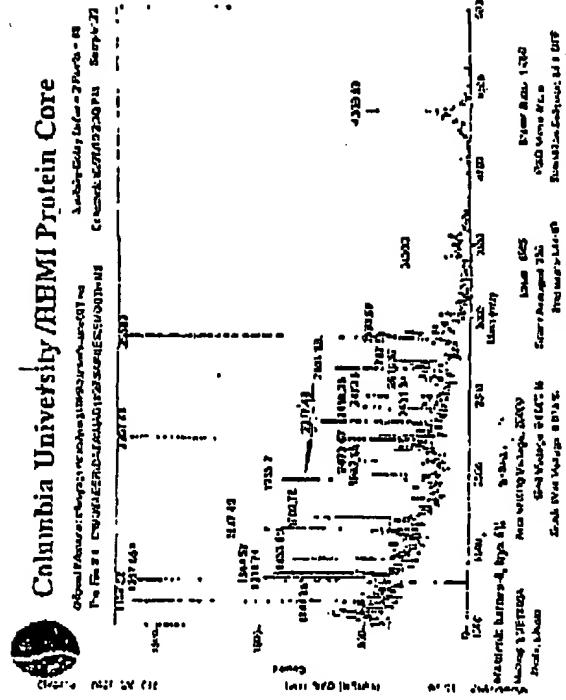
Figure 7B ~~2~~ (Band 2)



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Figure 7E (Band 5)

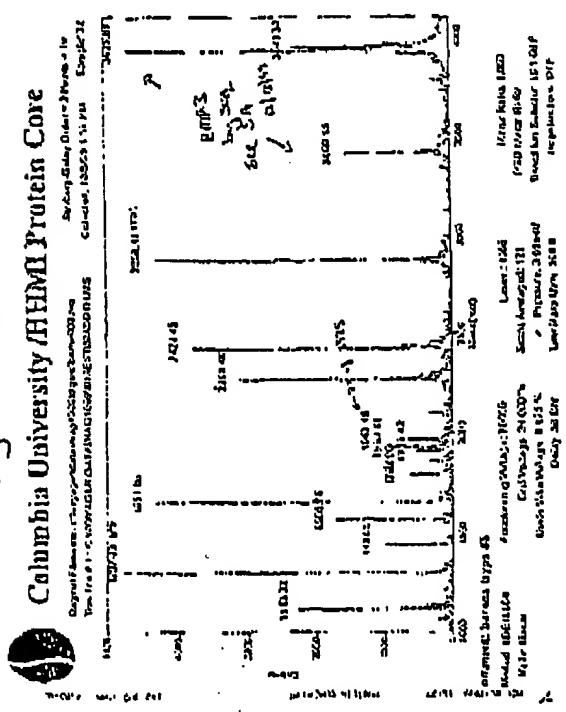
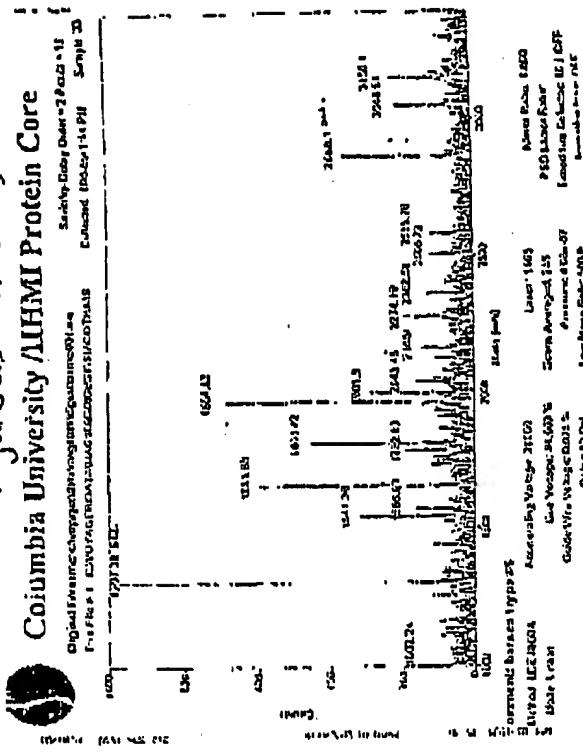
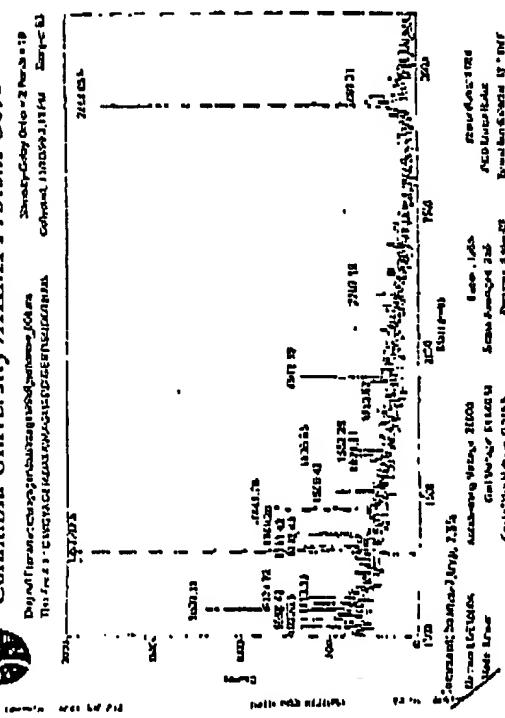


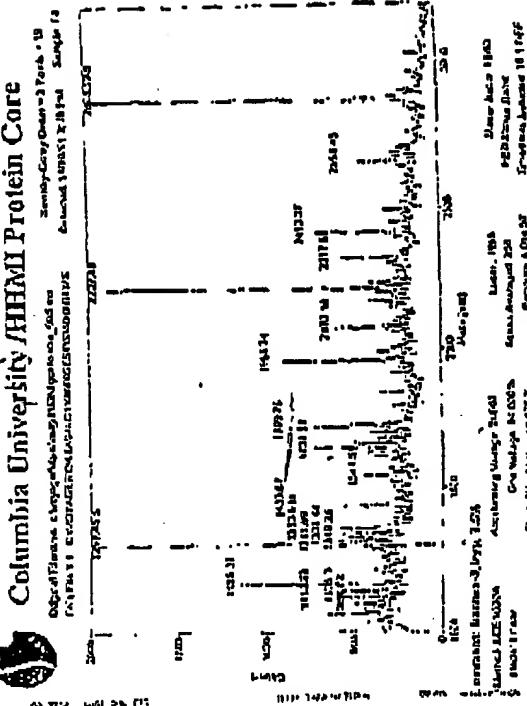
Figure 1 (continued)



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Figure ~~7~~ (Band 7)

Figure ~~7~~ (Band a)

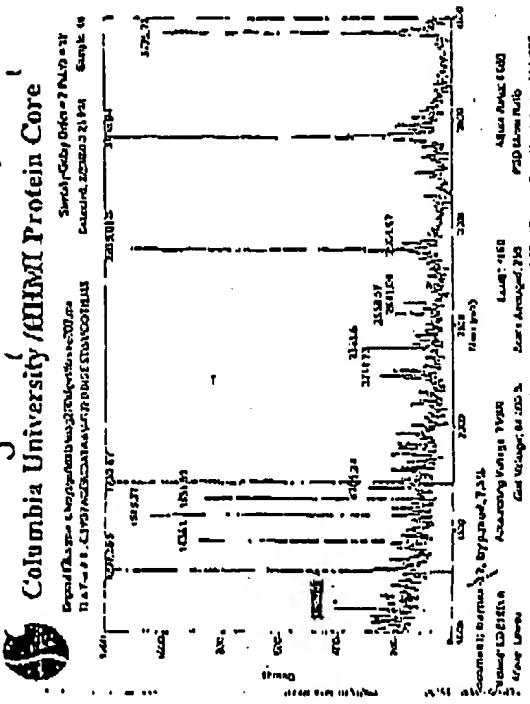
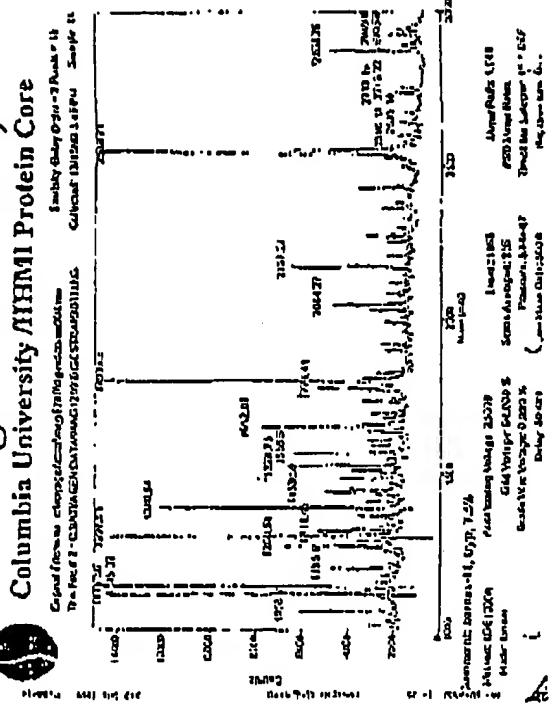
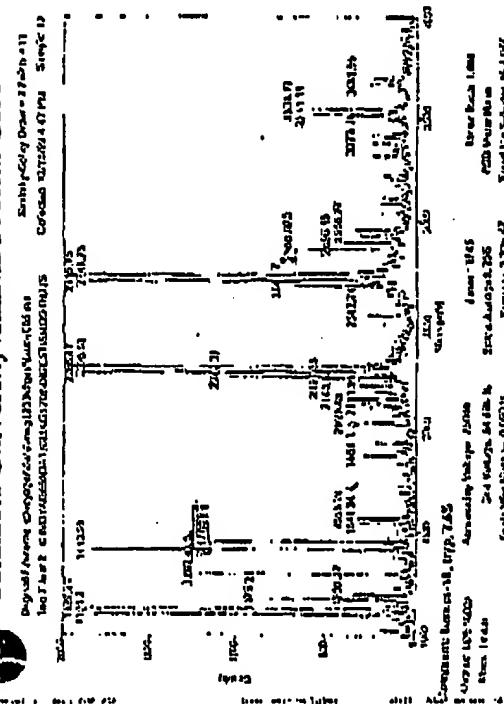


Figure 7.J (Band II)



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Figure 7M (Band 22)

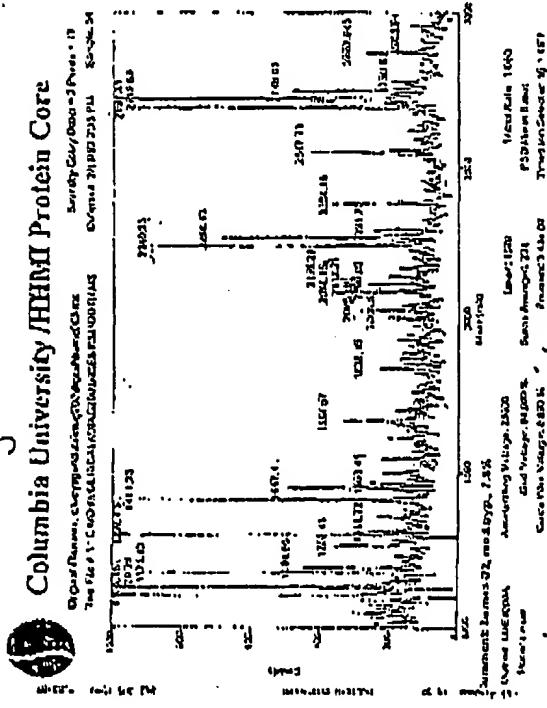


Figure 7N (Band 25)

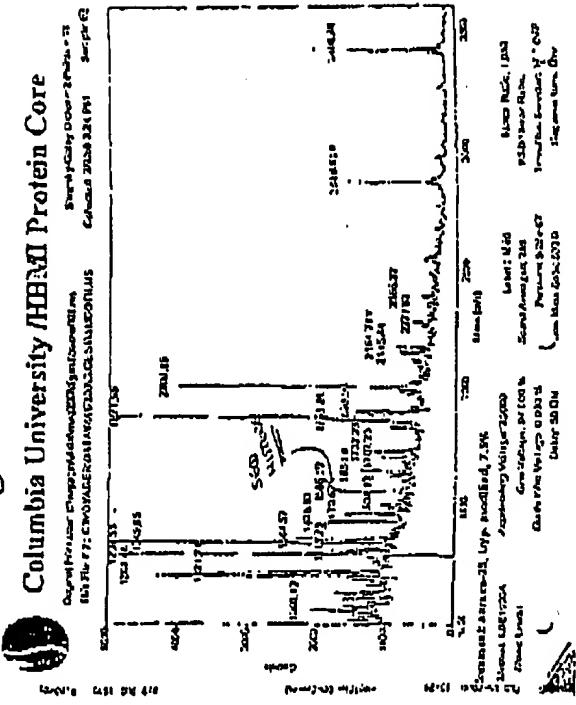


Figure 70 (Band 29)

FIGURE 8



00227-2502750

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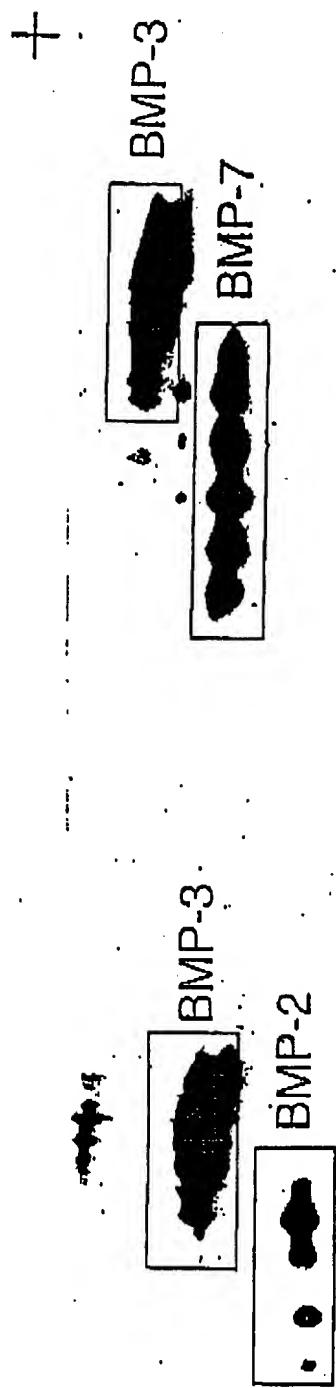


FIGURE 9A

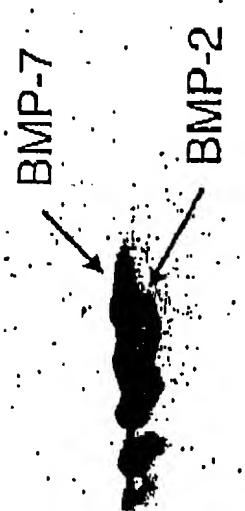


FIGURE 9B



FIGURE 9C



FIGURE 9D

52

FIGURE 10

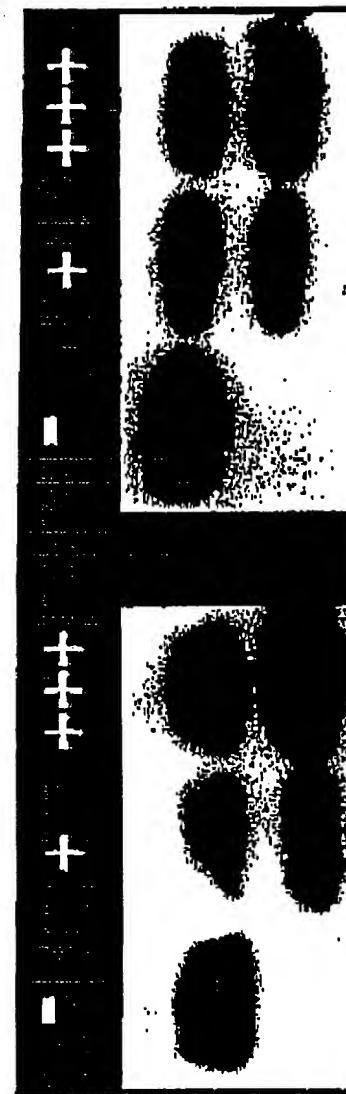
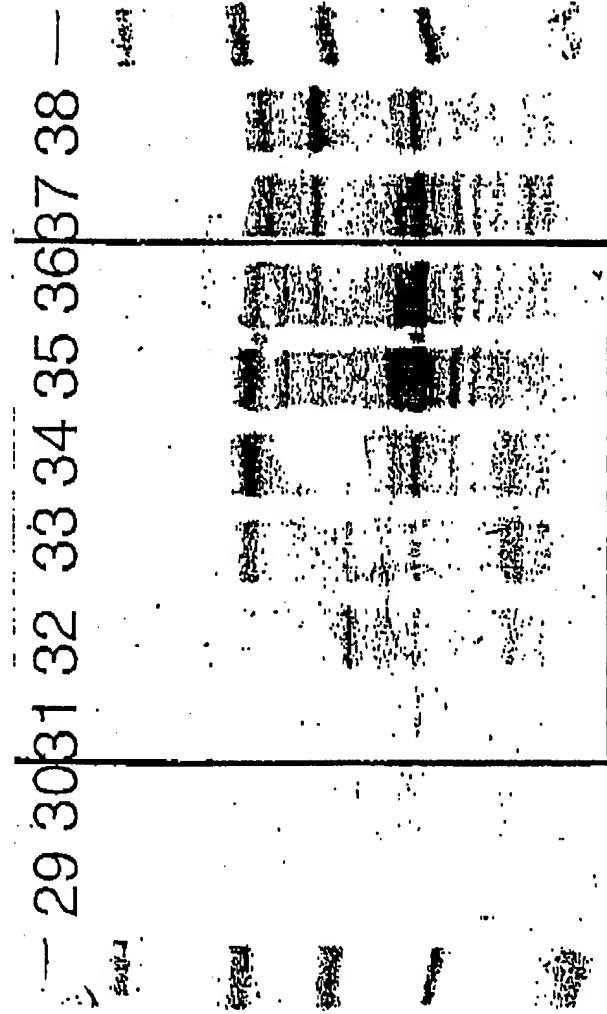


FIGURE 11

FIGURE 12

FIGURE 13A

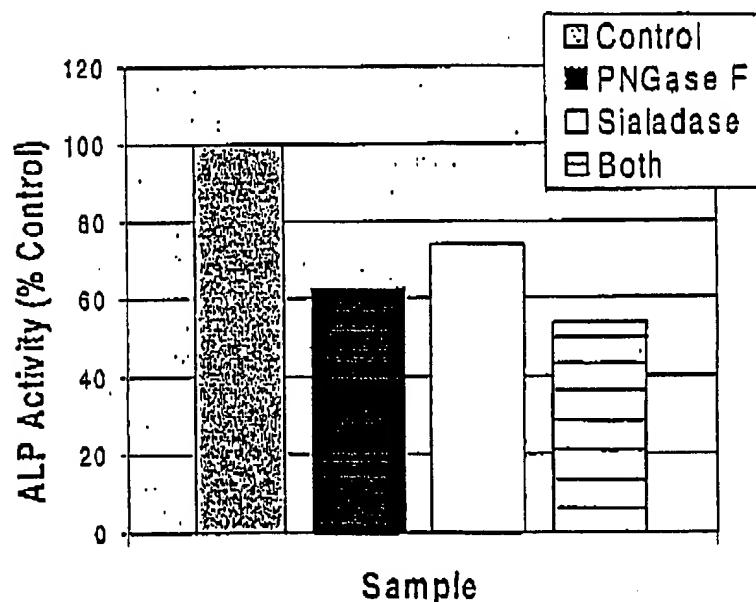
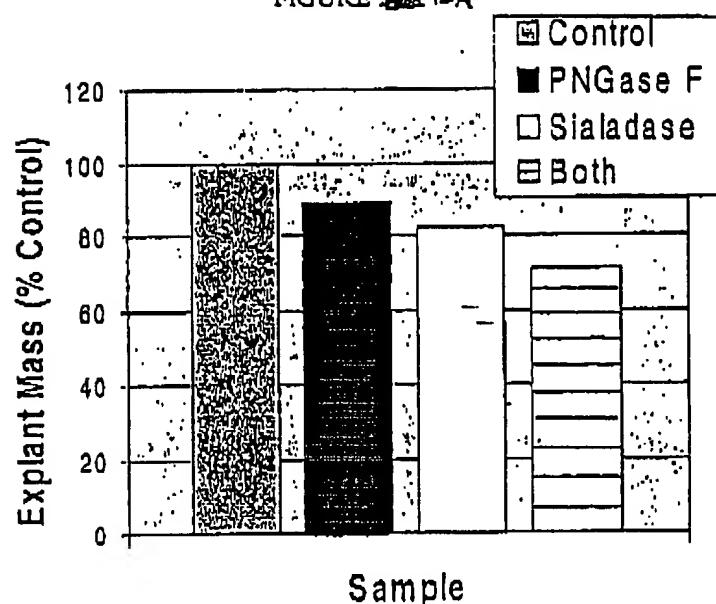


FIGURE 13B

Figure 14 Antibody Listing

Specificity	Antigen	Host Species	PC/NC	Source	Catalog No.
TGF- β 1 (human)	Protein	Rabbit	Polyclonal	Promega	GI1221
TGF- β 2 (human)	Peptide	Rabbit	Polyclonal	Santa Cruz Biotechnology	sc-90
TGF- β 3 (human)	Peptide	Rabbit	Polyclonal	Santa Cruz Biotechnology	sc-82
BMP-2 (human)	Protein	Rabbit	Polyclonal	Austral Biologics	PA-513-9
BMP-3 (human)	Peptide	Chicken	Polyclonal	Research Genetics	NA
BMP-4 (human)	Peptide	Goat	Polyclonal	Santa Cruz Biotechnology	sc-6896
BMP-5 (human)	Peptide	Goat	Polyclonal	Santa Cruz Biotechnology	sc-7405
BMP-6 (human)	Peptide	Mouse	Monoclonal	Novocastra Laboratories	NCL-BMP6
BMP-7 (human)	Peptide	Rabbit	Polyclonal	Research Genetics	NA
TGF- β 1 (human)	Peptide	Goat	Polyclonal	Santa Cruz Biotechnology	sc-1884
osteonectin (bovine)	Protein	Mouse	Monoclonal	DSHB	AON-1
osteocalcin (bovine)	Protein	Rabbit	Polyclonal	Accurate Chemicals	A761/R1H
serum albumin (bovine)	Protein	Rabbit	Polyclonal	Chemicon International	AB870
transferrin (human)	Protein	Chicken	Polyclonal	Chemicon International	AB797
apo-A1 lipoprotein (human)	Protein	Goat	Polyclonal	Chemicon International	AB740

Figure 15A Identification of Proteins by Amino Acid Sequencing of Tryptic Fragments from 1D Gels

Band	Sample	Sequence Data	Best Database Match	Match	Identification	Species	Acc. No.	Ref.
1								
2	fx 49 (1579)	XLAAGYDVEK	ALAAAGYDVEK	11/11	histone H1.c	human	676668 (NCBI)	65-75
3	fx 67 (1346)	SEKVCADLR	SEKVCADLR	11/11	40s Ribosomal Protein S20	rat	R3RT2D (PIR)	31-41
4	fx 65 0 (1580)	(V)YCCGMLGFSEAPV	WVCGMLGFGEKRV	11/14	LORP	mouse	AAC953301 (NCBI)	213-228
5	N terminal seq	STGVILPLQNNELPG	STGVILPLQNNELPG	15/15	BMP-3	human	4557371 (NCBI)	290-304
fx 72 (3425)	STGVILPLQNNELPG EYQY	STGVILPLQNNELPG AEYQY	20/20	BMP-3	human	4557371 (NCBI)	290-309	
fx 74 (3409)	STGVILPLQ	STGVILPLQ	9/9	BMP-3	human	4557371 (NCBI)	290-298	
6	fx 55 (1506)	(S)QTLQFQE	SATLQFDE	7/8	BMP-3	human	4557371 (NCBI)	346-353
fx 47	WYAF	no match		1/1?				
N terminal seq	HAGKYSREKNT(P)AIP	[HGGKYSREKNT(P)K]	11/14	α -2-Macroglobulin Receptor Assoc. Pro.	human	P30533 (Swiss-Pro)	31-46	
fx 57 (1438)	SQTLQFDEQ	SQTLQFDEQ	9/9	BMP-3	human	4557371 (NCBI)	346-354	
fx 57 (1852)	SLKPSNH-A	SLKPSNH-A	8/8	BMP-3	human	4557371 (NCBI)	410-417	
7	fx 51 (1553)	AALRPLVXP	AALRPLVXP	9/9	60s Ribosomal Protein L32	mouse	P17832 (Swiss-Pro)	1-9
fx 37 (no MS)	A(H)(Q)IVERYV	AIVER	5/5	60s Ribosomal Protein L32	mouse	P17832 (Swiss-Pro)	109-113	
fx 37 (no MS)	A(H)(Q)IVERYV	HQ5DRVV	5/7	60s Ribosomal Protein L32	mouse	P17832 (Swiss-Pro)	22-28	
8	fx 78 0 (1587)	XALFG(A)DQGXALGP	no match	7/7				
9	fx 58 (1587)	SQTLQFDEQT	SQTLQFDEQT	10/10	BMP-3	human	P12645 (Swiss-Pro)	346-355

Figure 15B Identification of Proteins by Amino Acid Sequencing of Tryptic Fragments from 1D Gels

Band	Sample	Sequence Data	Best Database Match	Match	Identification	Species	Acc. No.	AAs
11	fx 55 (1311)	SQTLXF	SQTLQF	5/6	BIIMP-3	human	43557371 (NCBI)	346-351
	fx 47 (1772)	VLATVTKPVGDK	VLATVTKPVGGDK	13/13	60s Ribosomal Protein L6	human	Q02878 (Swiss-Prote)	87-93
	fx 76 (1785)	IVFAL		4/4	60s Ribosomal Protein L6	human	Q02878 (Swiss-Prote)	273-276
	fx 51 (1145)	AVPQLQGTYLR	AIPOLOGTYLR	9/10	60s Ribosomal Protein L6	human	Q02878 (Swiss-Prote)	267-271
18								
22	fx 58 (1101)	ALDAAYYCFR	ALDAAYYCFR	9/9	TGF- β 2	human	P08112 (Swiss-Prote)	303-311
	fx 69 (no match)	GYNANFCAGACPYL	GYNANFCAGACPYL	14/14	TGF- β 2	human	P08112 (Swiss-Prote)	340-353
	fx 66 (1411.71)	VNSQSLSPY	VNSQSLSPY	9/9	SPP24	bovine	Q27967 (Swiss-Prote)	42-50
25	fx 39 (1470)	KAAKPSV(P)	KAAKPSV(P)	8/8	Histone H1.x	human	JCA926 (PIR)	199-206
29								

fx = fraction number (molecular weight of fragment, as measured by SDS-PAGE)

Figure 14A Identification of Proteins by Mass Spectrometry of Tryptic Fragments from 1D Gels

Band	Mass Spec Profile	Species	Acc. No.	Mass Spec Data	Mass Database	Mass Difference	AAS	% Coverage	Comments
1	4 peaks match with histone H1.c	human	87668B (NCBI)	1172.97	1172.37	0.60	110-121	22	15 MS peaks match with Band 2
				1570.87	1570.71	0.16	55-78		
				1708.47	1707.89	0.58	64-78		
				2011.58	2012.32	-0.74	35-54		
2	3 peaks match with histone H1.c	human	87668B (NCBI)	1570.76	1570.71	0.05	65-79	16	Identification of stained peptide confirmed by sequence analysis
				1708.02	1707.89	0.13	64-79		
				2012.12	2012.32	-0.20	35-54		
3	7 peaks match with ribosome S20	rat	R3R120 (PfR)	1129.76	1129.40	0.36	50-59	62	
				1158.21	1156.30	-0.09	76-83		
				1334.46	1334.62	-0.16	58-66		
				1352.13	1351.58	0.55	88-99		
				1518.04	1517.77	0.27	9-21		
				1919.02	1919.19	-0.17	5-21		
				3404.02	3404.87	-0.85	88-119		
4	3 peaks match with Lysyl Oxidase RP	human	NP002349 (Swiss-Pro)	1937.95	1938.27	-0.32	150-167	8	12 MS peaks match with Band 8
				2410.35	2410.63	-0.28	64B-66B		
				2610.57	2610.10	0.47	455-478		

Figure 16B Identification of Proteins by Mass Spectrometry of Tryptic Fragments from 1D Gels

Band	Mass Spec Profile	Species	Acc. No.	Mass Spec Data	Mass Spec Database	Mass Difference	A45	% Coverage	Comments
5	9 peaks match with BMP-3	human	4557371 (NCBI)	1113.32	1113.31	0.01	361-363	48	% coverage calculation is relative to the mature BMP-3, 183 AAS (290-472)
				1438.53	1438.58	-0.05	346-357		
				1566.76	1566.76	0.00	345-357		
				1651.86	1651.91	-0.05	410-424		
				1794.09	1794.02	0.07	346-380		
				2268.46	2268.53	-0.17	374-392		
				2424.45	2424.181	-0.36	373-392		
				3409.15	3407.77	1.38	290-318*		
				1002.24	1002.15	0.09	283-290	17	
				P30533 (Swiss-Pro)					
6	3 peaks match with $\alpha 2$ -Macroglobulin RAP	human		2362.58	2362.43	0.15	129-150		Identification of stained peptide confirmed by sequence analysis
				3048.51	3048.52	-0.01	257-282		
				1566.93	1566.75	0.18	346-357	15	
7	2 peaks match with BMP-3	human	4557371 (NCBI)	1651.88	1651.91	-0.03	410-424		% coverage calculation is relative to the mature BMP-3, 183 AAS (290-472)

Figure 16C Identification of Proteins by Mass Spectrometry of Tryptic Fragments from 1D Gels

Band	Mass Spec Profile	Species	Acc. No.	Mass Spec Data	Mass Database	Mass Difference	AAs	% Coverage	Comments
7	4 peaks match with ribosome L32	mouse	P17932 (Swiss-Pro)	1033.25	1033.17	0.08	67-75	33	
				1093.31	1093.40	-0.09	1-10*		
				1134.72	1134.28	0.44	65-74		
				1440.78	1449.68	0.12	19-29		
	5 peaks match with BMP-3	human	4557371 (NCBI)	1060.42	1060.20	0.22	102-111	21	% coverage calculation is relative to the mature BMP-3, 183 AAs (290-472)
				1113.39	1113.31	0.08	361-368		
				1380.28	1360.58	-0.32	190-210		
				1652.28	1651.91	0.37	410-424		
				1793.62	1794.02	-0.40	346-360		
B	1 peak matches with Lysyl Oxidase RP	human	NP002309 (Swiss-Pro)	2410.37	2410.63	-0.26	648-659	3	12 MS peaks match with Band 4
9	6 peaks match with BMP-3	human	4557371 (NCBI)	1113.14	1113.31	-0.17	361-368	36	% coverage calculation is relative to the mature BMP-3, 183 AAs (290-472)
				1438.60	1438.58	0.02	346-357		
				1586.77	1558.76	0.01	345-357		
				1651.91	1651.61	0.30	410-424		
				2901.67	2901.19	0.48	41-66		
				3408.94	3407.77	1.17	290-318		

Figure 16D Identification of Proteins by Mass Spectrometry of Tryptic Fragments from 1D Gels

Band	Mass Spec Profile	Species	Acc. No.	Mass Spec Data	Mass Spec Database	Mass Difference	ΔLs	% Coverage	Comments
11	5 peaks match with BMP-3	human	4557371 (NCBI)	1113.23	1113.31	-0.08	361-368	48	% coverage calculation is relative to the mature BMP-3, 183 AAS (290-472)
				1651.73	1651.91	-0.18	410-424	44	
				1793.58	1794.02	-0.44	348-380	32	
				2424.24	2424.81	-0.57	373-392	21	
				3408.34	3407.77	0.57	280-318	16	
12	5 peaks match with ribosome L6	human	Q02878 (Swiss-Prote)	1140.38	1140.23	0.15	114-122	16	
				1526.88	1526.86	0.02	141-155	10	
				1059.15	1059.12	0.03	10-20	10	
				1145.38	1145.35	0.01	262-271	9	
				1386.74	1386.68	0.06	280-271	8	
13	4 peaks match with TGF-β2	human	P08112 (Swiss-Prote)	1101.20	1101.26	-0.06	303-311	52	
				1175.26	1175.42	-0.16	400-409	40	
				2240.37	2240.80	-0.23	312-320	30	
				2691.70	2691.91	-0.21	340-362	28	
				1410.93	1411.60	-0.67	42-53	30	
14	5 peaks match with SPP24	bovine	Q27797 (Swiss-Prote)	1447.59	1447.65	-0.06	113-124	12	
				1540.84	1540.80	0.04	88-98	10	
				1869.10	1869.05	0.05	62-77	9	
				2268.47	2268.57	-0.10	33-53	13	

Figure 16E Identification of Proteins by Mass Spectrometry of Tryptic Fragments from 1D Gels

Band	Mass Spec Profile	Species	Spec No.	Acc. No.	Mass Spec Data	Mass Database	Mass Difference	AAs	% Coverage	Comments
22	5 peaks match with TGF- β 2	human	P08112 (Swiss-Prot)	1101.15	1101.26	-0.11	303-311	63		
				1175.13	1175.42	-0.29	400-409			
				2084.18	2084.42	-0.26	312-347			
				2240.25	2240.60	-0.35	312-328			
				2691.61	2691.91	-0.30	340-362			
22	2 peaks match with SPP24	bovine	Q27867 (Swiss-Prot)	1411.23	1411.60	-0.37	42-53	11		
				1447.40	1447.65	-0.25	113-124			
25	5 peaks match with histone H1x	human	JC4828 (PIR)	1208.46	1208.40	0.06	48-57	14		
				1221.71	1222.35	-0.64	107-118			
				1349.85	1350.52	-0.67	107-119			
				1384.57	1384.59	-0.02	48-58			
				1732.23	1732.97	-0.74	43-57			
5 peaks	match with BIMP-3	human	4557374 (NCBI)	1050.43	1050.20	0.23	102-111	31	% coverage calculation is relative to the mature BIMP-3, 183 AAs (290-472)	
				1438.83	1438.58	0.25	346-357			
				1566.92	1568.76	0.16	345-357			
				1651.80	1651.91	-0.11	410-424			
				3408.86	3407.77	1.09	290-318			

Figure 16F Identification of Proteins by Mass Spectrometry of Trypsic Fragments from 1D Gels

Band	Mass Spec Profile	Species	Acc. No.	Mass Spec Data	Mass Database	Mass Difference	AAs	% Coverage	Comments
29	4 peaks match with BMP-3	human	4557371 (NCB)	1113.22	1113.31	-0.09	381-368	27	% coverage calculation is relative to the mature BMP-3, 1B3 AAS (29D-472)

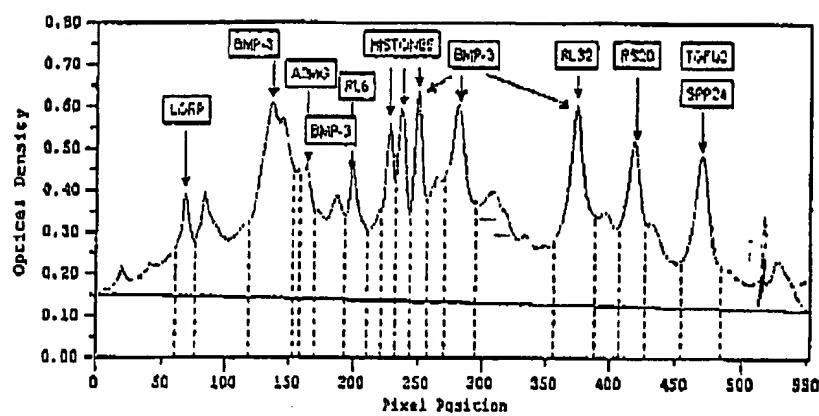


FIGURE 17A



FIGURE 17B

Figure 18 Quantitation of Identified BP proteins

Identified Protein	Percentage of Total Protein
LORP	2
BMP-3	11
BMP-3 & A2-MG	3
RL6 & BMP-3	4
Histone	3
Histone	3
Histone & BMP-3	4
BMP-3	8
RL32 & BMP-3	8
RS20	5
SPP24 & TGF- β 2	6
Total	58%

Figure 19A Identification of Protein by Mass Spectrometry of Fragments from 2D Gels

Spot	Digest	Mass Spec Profile	Species	Acc. No.	MS Peaks		AAS	% Coverage	Comments
					Data	Database			
1	Lys-C	2 peaks match with Coagulation Factor XIIIa	Human	P05160 (Swiss-Proj)	1837.01	1837.14	-0.13	472-487	8
					1921.65	1921.14	0.51	388-392	
					2679.51	N/A	N/A	488-504	peptide match confirmed by sequence analysis
2	Trypsin	2 peaks match with LORP	Human	NP002309 (Swiss-Proj)	1609.57	1609.88	-0.31	241-253	5
					2410.68	2410.63	0.26	648-668	
3	Lys-C	8 peaks match with Cathepsin L Precursor	Bovine	P25975 (Swiss-Proj)	1407.26	1406.80	0.46	105-116	41
					1546.84	1546.70	0.14	56-70	
					1681.16	1680.80	0.36	21-33	
					1681.86	1680.80	1.06	301-314	
					1834.71	1834.60	0.71	318-334	
					2352.90	2351.50	1.40	274-285	
					2381.50	2380.70	0.80	239-261	
					2721.51	2721.10	0.41	131-154	

Figure 7C Identification of Proteins by Mass Spectrometry of Fragments from 2D Gels

Spot	Digest	Mass Spec Profile	Species	Acc. No.	MS Peaks		AAS		% Coverage	Comments
					Data	Database	Diff			
7	Lys-C	4 peaks match with TGF- β 2	Bovine	P21214 (Swiss-Prote)	774.56	774.90	-0.34	26-31	42	
					809.69	809.94	-0.25	32-37		
					1175.12	1175.43	-0.31	88-107		
					3168.10	3168.66	1.44	1-25		
					2167.77	2167.51	0.26	42-60	10	
		1 peak matches with SPP24	Bovine	Q227967 (Swiss-Prote)						
	B	Trypsin	12 peaks match with ribosome L3	Bovine	P369872 (Swiss-Prote)	917.39	917.14	0.25	348-355	37
					984.23	984.15	0.08	10-18		
					1192.52	1192.40	0.22	286-296		
					1380.67	1380.65	0.02	249-260		
					1464.80	1464.63	0.17	103-114		
					1620.86	1620.82	0.04	103-115		
					1778.94	1778.00	-0.16	34-49		
					2238.43	2238.55	-0.12	30-49		
					2325.98	2325.65	0.34	177-197		
					2661.31	2661.04	0.27	220-223		
					2699.94	2698.43	-0.49	70-93		
					2946.10	2946.35	-0.25	198-223		

Figure 7/10 Identification of Proteins by Mass Spectrometry of Fragments from 2D Gels

Spot	Digest	Mass Spec Profile	Species	Acc. No.	MS Peaks Data	Database Data	Diff	AAS	% Coverage	Comments
9	Trypsin	7 peaks match S3a	Mouse	P97351 (Swiss-Proj)	920.05	920.10	-0.05	19.26	28	
					1218.28	1218.31	-0.02	152.161		
					1346.62	1346.49	0.13	151.161		
					1516.69	1516.69	0.00	174.185		
					1593.72	1593.82	-0.10	94.108		
					1719.91	1720.00	-0.09	199.212		
					1953.12	1953.16	-0.04	63.51		
10	Trypsin	4 peaks match H1.c	Human	87668 (NCBI)	1327.75	1327.58	0.19	34.46	23	
					1579.70	1579.71	-0.01	65.79		
					1707.65	1707.89	-0.24	64.79		
					2147.17	2147.53	-0.38	1.21		
11	Trypsin	6 peaks match S4	Human	P12750 (Swiss-Proj)	1188.48	1168.38	0.10	230.239	1 23	
					1216.38	1216.39	0.00	134.144		
					1354.03	1353.61	0.42	230.241		
					1507.81	1507.88	0.12	198.210		
					1557.75	1557.98	-0.23	37.48		
					2140.34	2140.58	-0.24	221.239		
					2591.80	2591.90	-0.10	77.98		